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DEVELOPMENT OF AN UPLC-MS/MS METHOD FOR THE DETERMINATION OF SEVEN UREMIC RETENTION TOXINS IN SERUM OF CHRONIC KIDNEY DISEASE PATIENTS

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Chronic kidney disease (CKD) is a devastating illness characterized by the accumulation of uremic retention solutes in the body. The objective of this study was to develop a simple and robust UPLC-MS/MS method for the simultaneous determination of seven organic acid uremic retention toxins in serum, namely uric acid (UA), hippuric acid (HA), indoxylsulfate (IS), p-cresylglucuronide (pCG), p-cresylsulfate (pCS), indole-3-acetic acid (IAA) and 3-carboxy-4-methyl-5-propyl-2-furanpropionic acid (CMPF). Isotopically labelled internals standards (d_5-HA; 1,3-$^{15}N_2$-UA and d_5-IAA) were used to correct for sample preparation and system performance variations. Separation was performed on a C18 column followed by negative electrospray ionization and tandem mass spectrometric detection. The within-day precision varied from 0.60 to 4.54% and the between-day precisions were lower than 13.32% for all compounds. The matrix effect was less than 15%. Accuracy ranged from -0.34 to 3.05% for UA, -0.89 to -9.67% for HA, -0.31 to -4.98% for IS, -0.98 to -11.80% for pCG, 1.40 to 11.00% for pCS, -0.67 to -12.33% for IAA and 2.00 to 14.84% for CMPF. The applicability of the method was evaluated by analyzing 78 serum samples originating from healthy controls as well as from patients in the different stages of CKD. These results were compared with those obtained by commonly used HPLC-PDA-FLD methods. A good correlation was obtained between the methods for all analytes.

Keywords: chronic kidney disease, uremic toxins, UPLC-MS/MS